Invited Review

CROI 2016: Complications of HIV Infection and Antiretroviral Therapy

Diane V. Havlir, MD; Judith S. Currier, MD

Noncommunicable conditions such as cardiovascular disease, hypertension, renal and bone diseases, and malignancies as well as infectious complications are an ongoing concern during the course of treated HIV disease. Research in this area continues to focus on the epidemiology and risk factors for these conditions, on identifying the contributions of HIV-related immunopathology to specific and collective end-organ diseases, and on evaluating interventions to prevent or reduce the morbidity associated with these conditions. Data presented at the 2016 Conference on Retroviruses and Opportunistic Infections provided new insights into all of these areas.

Keywords: CROI, 2016, complications, HIV, cardiovascular disease, comorbidities, statins, bone, renal, pulmonary, malignancy, tuberculosis, opportunistic infections, cryptococcal meningitis, non–AIDS-related events, atherosclerosis, CVD, CVD risk, adipose tissue, cancer, resistance, screening

New information on the long-term complications of HIV disease remained a major focus at the 2016 Conference on Retroviruses and Opportunistic Infections (CROI), including studies focused on noncommunicable chronic diseases (eg, cardiovascular, renal, and bone diseases) and opportunistic infections (eg, *Mycobacterium tuberculosis* and *Cryptococcus* infections). Progress in identifying predictors of specific complications as well as interventions for their prevention and treatment are reviewed below.

Non–AIDS-Related Events

With the current goal of viral suppression for all people living with HIV infection, new tools are needed to identify those who remain at risk for serious non–AIDS-related events, so that strategies to reduce these complications can be targeted. Previous studies have demonstrated a relationship between lower CD4+/CD8+ cell ratio and the risk for morbidity and mortality caused by non–AIDS-related events.[1] McGettrick and colleagues examined risk factors for non–AIDS-related events among a cohort of individuals taking antiretroviral therapy in Ireland and found that older age at initiation of therapy, injection drug use, and preevent CD4+/CD8+ cell ratio ere independently associated with risk for non–AIDS-related events (Abstract 710). The researchers were able to identify an incremental increase in risk for non–AIDS-related events across a range of CD4+/CD8+ cell ratios; for those with a CD4+/CD8+ cell ratio below 0.26, the hazard ratio (HR) for an event was 3.11 compared with nonsignificant associations for those with ratios above this level. Although options beyond suppressive antiretroviral therapy to raise the CD4+/CD8+ cell ratio are currently limited, these thresholds could be used to target other interventions to reduce non–AIDS-related events in future studies.

Cardiovascular Disease

An analysis of the North American AIDS Cohort Collaboration on Research and Design (NA-ACCORD) cohort provided more evidence that HIV-infected adults have a higher risk of myocardial infarction (MI) than uninfected controls (Abstract 641). In this analysis, rates of type 1 MI among 7 cohorts of HIV-infected adults in NA-ACCORD were compared with the diverse population of presumably uninfected adults followed in the MESA (Multi-Ethnic Study of Atherosclerosis) and the ARIC (Atherosclerosis Risk in Communities) studies. Adjusted incidence rate ratios for MI in the HIV-infected group ranged between 1.3 (ARIC) and 2.4 (MESA) times higher than in uninfected control groups, after controlling for demographic factors and smoking status but not hypertension or dyslipidemia. These results confirm prior studies that demonstrated an excess risk of MI among adults with HIV infection, using careful control for demographic factors.

The optimal measure for predicting the risk of cardiovascular disease (CVD) remains controversial. This topic was addressed by several well-designed studies (Abstracts 42, 641, 642, and 643). Crane and colleagues used prospectively collected data from the Center for AIDS Research (CFAR) Network of Integrated Clinical Systems (CNICS) cohort to compare the performance of 4 CVD risk scores: 1) Framingham risk score; 2) Adult Treatment Panel (ATP) III risk score; 3) 2013 American College of Cardiology (ACC)/American Heart Association (AHA) atherosclerotic CVD (ASCVD) risk score; and 4) the HIV-specific CVD risk score from the D:A:D (Data Collection on Adverse Events of Anti-HIV Drugs) study (Abstract 42). The

Dr Havlir is Professor of Medicine at University of California San Francisco and Chief of the HIV, Infectious Diseases, and Global Medicine Division at San Francisco General Hospital. Dr Currier is Professor of Medicine and Chief of the Division of Infectious Diseases and Associate Director of the Clinical AIDS Research and Education (CARE) Center at University of California Los Angeles. Send correspondence to Diane V. Havlir, MD, San Francisco General Hospital, 995 Potrero Avenue, UCSF Box 0872 San Francisco, CA 94110. Received on March 17, 2016; accepted on April 6, 2016.

investigators also examined the prediction of different types of MI (type 1 due to atherosclerotic plaque and type 2 due to oxygen supply/demand mismatch). In this very large study of 11,338 people with 243 incident MIs, the investigators found that the 2013 ACC/AHA ASCVD risk model performed as well or better than other risk scores across all MI events and that this score outperformed the others for predicting type 2 MI events.

Clement reported on CVD risk prediction using data from the Veterans Affairs Clinical Case Registry that included 3171 male veterans with follow-up over a 10-year period (Abstract 642). In this study, the end points included a broader range of CVD events beyond MI. In contrast to the NA-ACCORD study, the D:A:D model performed better than the ACC/AHA ASCVD risk calculator, and inclusion of hepatitis C virus infection and HIV RNA into a new model improved discrimination.

Weigel compared the 2013 ACC/AHA guidelines[2] on ASCVD risk with the 2004 ATP III recommendations[3] in 352 individuals without prior ASCVD who had available data on the presence of carotid plaque and 3-year rates of progression of carotid intima-media thickness (CIMT), to determine whether the guidelines recommended the use of statins in those with evidence of disease. The investigators found that although the 2013 ACC/AHA guidelines recommended statins to a greater percentage of individuals with carotid plaque, the 2004 guidelines better correlated with individuals with progression of CIMT. Whether the inclusion of a measure of subclinical atherosclerosis such as CIMT would improve longer-term risk prediction remains to be determined. Further improvements in HIV risk prediction can likely be obtained with fine tuning of the available risk equations. In the interim, the use of the 2013 ACC/AHA ASCVD risk score for predicting MI is unlikely to overestimate CVD risk.

The role of noninvasive cardiovascular testing to stratify coronary artery disease risk among individuals with HIV infection also remains poorly defined. Feinstein and colleagues (Abstract 644) used a clever approach to obtain data on this question by reviewing the outcome of stress test results within the Northwestern University database. Among individuals with a positive stress test result, those with HIV infection were more likely to have a greater burden of coronary artery disease during angiography and a higher rate of percutaneous coronary interventions. Whether this represents a difference in testing rates or a higher rate of subclinical disease awaits further study.

Further improvements in HIV risk prediction can likely be obtained with fine tuning of the available risk equations. In the interim, the use of the 2013 ACC/AHA ASCVD risk score for predicting MI is unlikely to overestimate CVD risk.

Biomarkers of Atherosclerosis and CVD Risk

Circulating biomarkers that predict the risk of MI or other measures of subclinical atherosclerosis help to reveal the pathogenesis of these problems and to identify potential targets for interventions as well as individuals at high risk. Numerous studies examined the relationship between a variety of plasma biomarkers and cellular markers and a range of different cardiovascular outcomes, including measures of endothelial function, CIMT, and clinical events (Abstracts 671-673 and 651-657). It is challenging to summarize and reconcile the findings from these studies, given the lack of standardized practices for controlling for other risk factors within each study and the fact that each type of end point could reflect a different mechanistic process. Despite this limitation, these findings highlight important insights into the pathogenesis of these clinical problems.

Hunt and colleagues found that along with C-reactive protein and D-dimer, higher levels of oxidized low-density lipoprotein (LDL) cholesterol were associated with MI (Abstract 671). Nou and colleagues demonstrated that the reduction in coronary artery plaque (as measured by coronary computed tomography angiography) observed during a clinical trial of atorvastatin therapy paralleled a reduction in oxidized LDL cholesterol, suggesting that the effect of the statin was mediated through this mechanism (Abstract 673). Collectively, these studies highlight the potential role of oxidized lipids in the pathogenesis of atherosclerosis in the context of HIV infection. In contrast, Kelesidis and colleagues did not find a statistically significant association between oxidized LDL and progression of CIMT in individuals who initiated antiretroviral therapy over 96 weeks (Abstract 672), possibly suggesting that the effects of oxidized lipids may not be evident in the CIMT measurement. Other factors that were associated with the risk of clinical MI events included higher levels of B-type natriuretic peptide (Abstract 646).

Immune Activation: Unique Roles for T Cells and Monocytes

Specific monocyte subpopulations (CD14+ + CD16+) and expression of surface markers have been associated with levels of plasma biomarkers of inflammation and with a higher risk for subclinical atherosclerosis.[4,5] Chow and colleagues (Abstract 652) reported an association between carotid bifurcation thickness and both the nonclassical monocyte population (CD14low/ + CD16+ +) and monocyte chemotactic protein–1 (MCP-1) and 2-year rates of coronary artery calcification in the Hawaii Aging With HIV study (Abstract 652). These results confirm the growing literature on the importance of innate immune activation and progression of atherosclerosis in HIV infection.

Crowe and colleagues previously showed that glucose metabolism within monocytes remains altered after antiretroviral therapy.[6] At CROI 2016, Palchaudhuri and colleagues demonstrated that higher expression of glucose transporter 1 (GLUT1) on intermediate proinflammatory monocytes (CD14+ + CD16+) was associated with higher levels of some plasma biomarkers of CVD risk (D-dimer and lower levels of high-density lipoprotein [HDL] cholesterol), suggesting that inhibitors of GLUT1 could be targeted to reduce inflammation mediated in treated HIV infection (Abstract 711).

Several studies examined the role of activated T cells among individuals with MI and in relation to measures of endothelial function and markers of endothelial activation (Abstracts 653, 654, and 655). Higher levels of CD8+ T cells expressing CC chemokine receptor 5 were observed prior to acute coronary syndrome, suggesting a possible therapeutic target (Abstract 655). When endothelial dysfunction was examined as the end point (as measured by flow-mediated dilation of the brachial artery), one small study reported an association between activated CD8+ T cells and flow-mediated dilation (Abstract 653), and another study noted stronger links between cytomegalovirus immunoglobulin G and cytomegalovirus-specific CD4+ T cells (Abstract 654). Measures of monocyte activation appeared to be more closely related to endothelial activation markers (intercellular adhesion molecule–1 [ICAM-1] and vascular cell adhesion molecule–1 [VCAM-1]), and T-cell activation was not related to this outcome measure. Taken together, these findings suggest that endothelial activation and vascular reactivity are influenced through different mechanisms. For a summary of the complex topic of immunopathogenesis of metabolic complications, see the webcast of the symposium talk by Crowe (Abstract 125).

Vascular Function and Antiretroviral Therapy

The degree to which early antiretroviral therapy reduces CVD risk remains unknown. Baker and colleagues compared vascular function between individuals with untreated HIV infection randomly assigned to early antiretroviral therapy (initiated at CD4+ cell counts >500/μL) in the START (Strategic Timing of Antiretroviral Treatment) study, by measuring radial artery waveforms with a tonometer (Abstract 41). The 332 participants in this substudy were young, predominantly men, and had low CVD risk at baseline. During a median follow-up period that included 30 months of antiretroviral therapy for the immediate-treatment group, there were no within-person or between-group differences in vascular function. These results suggest that among a population of young, healthy people, early antiretroviral therapy was not able to impact this specific measure of vascular function.

In contrast, Innes and colleagues examined a different measure of vascular function, aortofemoral pulse wave velocity (PWV; a measure of elevated arterial wall stiffness), in children who initiated antiretroviral therapy early in life compared with a demographically similar HIV-uninfected group (Abstract 658). The investigators demonstrated that PWV improved consistently among the HIV-seropositive group with a longer duration of antiretroviral therapy, whereas there were no changes in PWV with age in the control group. These results suggest that vascular function may improve over time with early initiation of antiretroviral therapy.

Interventions to Reduce Inflammation and CVD Risk

O'Brien and colleagues from the AIDS Clinical Trials Group (ACTG) performed a randomized controlled trial to follow up on an earlier uncontrolled observation that low doses of aspirin might reduce levels of innate immune activation in the setting of well-treated HIV infection (Abstract 44LB). In this trial, 100 mg and 300 mg doses of aspirin were compared with placebo during 12 weeks of treatment followed by a washout period. Although serum thromboxane was inhibited among those who received aspirin (suggesting participants were adherent), there was no impact of the aspirin intervention on soluble CD14 or on flow-mediated dilation of the brachial artery. In fact, one measure of monocyte activation (soluble CD163) rose in the group that received aspirin. The study population was not preselected for a group with elevated measures of immune activation at baseline. Hence, it is still possible that there may be subgroups for whom aspirin therapy may reduce inflammation, and this can hopefully be explored in future studies.

Cardiovascular Disease Among Women

CVD risk among women was the focus of a few studies at CROI 2016. Hanna and colleagues from the Women's Interagency HIV Study compared the use of interventions to reduce CVD risk among HIV-seropositive with that among HIV-seronegative women and found that HIV-seropositive women with hypertension and diabetes were more likely to be on treatment for these issues than were HIV-seronegative controls; however, a sizable fraction did not achieve target levels of control for either condition (Abstract 647).

Looby and colleagues used stored samples from a study that measured the burden of coronary artery plaque by computed tomography scan and examined the relationship between menopausal status, plaque burden, and measures of innate immune activation (Abstract 650). Using a novel marker of ovarian reserve, anti-Müllerian hormone (a measure of reproductive aging), they were able to show that lower levels of anti-Müllerian hormone were associated with higher plaque burden and with higher levels of innate immune activation. These results suggest a possible connection between reproductive aging and CVD risk in the context of HIV infection. These findings also highlight the need to prioritize CVD screening among HIV-infected women prior to the onset of clinical menopause.

Heart Failure

There is growing awareness of heart failure as an important clinical problem in the setting of treated HIV infection, including among children (Abstract 853). Factors that contribute to a decline in left ventricular function during HIV infection are poorly defined. Longenecker reported an association between the presence and quality of pericardial fat depots and left ventricular function in a study of 46 individuals that combined echocardiographic measures with findings on computed tomography scanning (Abstract 649). This novel finding of ectopic fat depots and diastolic function warrants further evaluation. Previous studies have demonstrated a link between depression and heart failure.[7] In a comprehensive study of US veterans, this link was confirmed for HIV-infected and uninfected men (Abstract 714).

Bone Disease

It is well established that bone mineral density (BMD) initially declines after the initiation of antiretroviral therapy, but the impact of this change on long-term risk for fracture and the contribution of specific antiretroviral drugs to fracture risk remain unclear. Borges and colleagues examined rates of fracture and femoral osteonecrosis among nearly 12,000 participants from the EuroSIDA study (Abstract 46). The investigators identified that well-known host factors such as age, race, and HIV disease status were associated with fracture risk; however, after controlling for these factors, use of tenofovir disoproxil fumarate (TDF) (but not duration of exposure) was an independent risk factor for fracture. The excess risk of fracture ranged from 25% for those who had ever received TDF to 40% for those currently taking TDF. This is one of the largest studies to date to quantify the relationship between TDF exposure and fracture risk.

Interventions to reduce fracture risk have included changing the antiretroviral regimen and the use of pharmacologic agents to restore bone density. Gallant and colleagues presented data from a clinical trial that randomly assigned individuals who were virologically suppressed on a regimen that contained emtricitabine and TDF to remain on the same regimen or to switch the TDF component to tenofovir alafenamide (TAF) (Abstract 29). After 48 weeks of follow-up, those who switched to the TAF-containing regimen remained virologically suppressed, and BMD increased in the group receiving TAF but declined in the group receiving TDF. Additionally, more participants in the group receiving TAF had an improvement of 3% or more in BMD at week 48. These results confirm that TAF may have more favorable effects than TDF on bone. Further study is needed to determine the clinical significance of these changes.

Ofotokun and colleagues examined the impact of a single dose of zoledronic acid, an injectable agent that inhibits osteoclast activity and reduces bone turnover, to prevent bone loss among individuals initiating antiretroviral therapy (Abstract 47). Antiretroviral therapy–naive participants who were beginning a regimen of ritonavir-boosted atazanavir plus TDF and emtricitabine were randomly assigned to receive a single dose of zoledronic acid 5 mg or a placebo and followed for 48 weeks. After 12 weeks of follow-up, bone loss (as measured by C-terminal telopeptide of collagen [CTx], a sensitive marker of bone resorption) was reduced by 74% in the group receiving zoledronic acid. The benefits of zoledronic acid were maintained over 48 weeks of follow-up, and the drug was well tolerated. If confirmed in larger studies, perhaps among patients receiving contemporary antiretroviral regimens, these findings highlight a promising approach to preventing bone loss when initiating antiretroviral therapy.

Injectable zoledronic acid reduced bone loss by 74% among individuals taking an antiretroviral regimen of ritonavir-boosted atazanavir plus TDF and emtricitabine.

There is also a need to identify the most effective strategies to prevent bone loss among youth who are being treated for HIV infection. In a randomized clinical trial that evaluated different dosage strategies for vitamin D treatment, Eckard and colleagues found that a monthly dose of vitamin D_3 60,000 IU was effective in improving BMD and reducing levels of markers of bone turnover in individuals aged 8 years to 26 years taking antiretroviral therapy with a baseline 25-hydroxyvitamin D level of 30 ng/mL or lower (Abstract 859).

Questions remain about the safety of maternal use of TDF and newborn BMD. Siberry and colleagues from the International Maternal, Pediatric, Adolescent AIDS Clinical Trials (IMPAACT) Network PROMISE (Promoting Maternal-Infant Survival Everywhere) study compared bone outcomes among infants born to pregnant women enrolled in a 3-arm randomized trial (Abstract 36). The trial compared an antiretroviral regimen of ritonavir-boosted lopinavir and TDF or zidovudine with maternal prevention of mother-to-child transmission (PMTCT) prophylaxis with zidovudine, single-dose nevirapine, and a "tail" of TDF and emtricitabine. Using whole-body and lumbar spine dual-energy X-ray absorptiometry (DXA) scanning, the investigators measured bone mineral content (BMC) by age 28 days at 8 African sites. Among the 425 infants with DXA data available, the investigators found no difference in BMC among those whose mothers were taking boosted lopinavir and TDF or zidovudine; however, levels of BMC were higher in infants exposed to the short-course PMTCT regimen than in those exposed to maternal 3-drug antiretroviral therapy. The clinical significance of this difference over the longer term remains to be identified. Overall, these results provide reassurance that maternal exposure to TDF does not lead to lower BMC than exposure to zidovudine, at least when combined with boosted lopinavir.

Further evidence that ritonavir-boosted lopinavir may play a role in bone loss was seen in a study of children in South Africa who were receiving boosted lopinavir and were randomly assigned to continue or to switch to an efavirenz-based regimen (Abstract 40). After an average of 2 years of follow-up, levels of BMC were lower in the children who remained on boosted lopinavir than in those who switched to efavirenz, even after controlling for several factors associated with BMC.

The use of oral preexposure prophylaxis (PrEP) containing emtricitabine and TDF has been associated with a reduction in BMD in men.[8] This is of particular concern for healthy young men who have not yet achieved peak bone mass. Grant and colleagues presented follow-up data from a bone density substudy of the iPrEx (Chemophrophylaxis for HIV Prevention in Men) study and the companion follow-up iPrEx OLE (iPrEx Open Label Extension) study (Abstract 48LB). In this report, the investigators examined changes in BMD among those participants with measureable levels of emtricitabine and TDF 24 weeks after treatment was discontinued and observed reversal of bone loss back to levels comparable to those in the placebo group over this period of time. The findings from this study confirm that bone loss

occurs during PrEP with emtricitabine and TDF (with drug levels needed for protection from HIV-1 infection) and that this loss reverses 6 months after treatment is discontinued. For a comprehensive review of the topic of bone disease and HIV infection, see the webcast of the symposium talk by Mallon (Abstract 126).

Renal Disease

A very large study from NA-ACCORD investigators examined changes in estimated glomerular filtration rate (eGFR) among individuals taking TDF compared with other nucleoside analogue reverse transcriptase inhibitors (nRTIs) and found that, among those with a baseline eGFR below 90 mL/min, renal function declined more quickly over the first 6 months of therapy with TDF than with other nRTIs but then recovered only in those taking TDF (Abstract 684). In contrast, no differences in renal function were observed between those using TDF and those using other nRTIs if the baseline eGFR was greater than 90 mL/min. These results confirm transient declines in renal function during initial antiretroviral therapy containing TDF among those with mild renal impairment.

TAF, a prodrug of tenofovir, achieves lower plasma levels of tenofovir than TDF with equivalent or higher levels of intracellular tenofovir diphosphate and, hence, is expected to cause fewer renal and bone adverse effects. Several studies evaluated the safety of TAF among individuals at risk for renal disease or with established renal impairment. Wohl and colleagues reported the results of 2 prospective studies that compared TAF-containing with TDF-containing initial antiretroviral regimens and examined changes in renal function in those with 2 or more risk factors for renal disease and in those without risk factors (Abstract 681). In individuals at risk for renal disease, a decline in renal function was less common among those who received TAF than those who received TDF.

In individuals at risk for renal disease, a decline in renal function was less common among those who received TAF than those who received TDF.

Safety data on the use of TAF through 72 weeks in patients with impaired renal function (eGFR 30-69 mL/min) were presented by Post and colleagues (Abstract 680). In this study, individuals with renal impairment were switched to the fixed-dose combination of elvitegravir, cobicistat, TAF, and emtricitabine. In addition to continued stabilization of eGFR, improvement in BMD and reductions in proteinuria and albuminuria were also observed. These data provide reassurance about the use of TAF to reduce the risk of developing renal dysfunction among individuals at high risk for renal disease as well as among those with preexisting renal impairment.

Adipose Tissue

Damouche and colleagues expanded on prior observations[9] describing a possible role of visceral fat as an anatomic location for HIV persistence and as a source of chronic inflammation (Abstract 271). In elegant studies using adult cynomolgus macaques (*Macaca fascicularis*) infected with simian immunodeficiency virus $(SIV)_{mac251}$ and uninfected animals as controls, the investigators provided evidence that lymphocytes and macrophages isolated from adipose tissue and the stromal vascular fraction of this tissue harbored SIV DNA and RNA even in the presence of suppressive antiretroviral therapy. These findings suggest that adipose tissue may play a role in both viral persistence and chronic immune activation and inflammation during HIV infection.

New information continues to emerge regarding the contributions of specific antiretroviral drugs to changes in fat. Data from a small randomized trial that included samples from subcutaneous fat biopsies demonstrated a greater reduction in mitochondrial DNA in adipose tissue among individuals who were randomly assigned to receive zidovudine combined with a nonnucleoside analogue reverse transcriptase inhibitor (NNRTI) or a protease inhibitor (Abstract 668). These results highlight the possible contributions of NNRTIs to fat loss.

Investigators from the European AIDS Treatment Network (NEAT) presented the results of a body composition substudy nested into a study of treatment with ritonavir-boosted darunavir combined with raltegravir (nRTI-sparing regimen) or TDF and emtricitabine (Abstract 45). No changes in limb fat were observed between treatment groups, but total body fat mass and mean trunk mass increased more in those who were taking the nRTI-sparing regimen. Whether this increase in fat mass resulted from the lack of nRTIs or from the use of raltegravir cannot be determined from this design. For a summary of the topic of visceral fat in the context of HIV infection, see the webcast of the symposium talk by McComsey (Abstract 123).

In summary, considerable progress is being made in deciphering the unique factors that contribute to the risk of several long-term complications of chronic diseases in the setting of treated HIV infection. Although it appears that earlier initiation of antiretroviral therapy may not have a major impact on the risk of some of these issues, more attention to screening and prevention of comorbidities along with improvements in antiretroviral therapy and the development of specific interventions to reduce immune activation will be needed to eliminate these conditions as a source of comorbidity globally.

Cancer

People living with HIV infection are at increased risk for human papillomavirus (HPV)-related cancer. Wilkin and colleagues presented data from the ACTG A5298 trial, a randomized, double-blind, placebo-controlled trial of the quadrivalent HPV vaccine among HIV-infected adults aged 27 years or older (Abstract 161). Five hundred seventy-five adults were enrolled in the trial, with a median age of 47 years; participants were followed up for a median of 2.6 years. A data and safety monitoring board stopped the study early for futility. The vaccine did not prevent persistent anal HPV

infections compared with placebo (13 vs 17, respectively; HR, 0.75; 95% confidence interval [CI], 0.45-1.26). In addition, the vaccine did not have an impact on the occurrence of anal high-grade squamous intraepithelial lesions (HSILs), the precursor to invasive anal cancer. Vaccinated participants experienced fewer persistent oral HPV infections than those who received a placebo (1 vs 8, respectively; HR, 0.22; 95% CI, 0.02-0.98). The vaccine was safe and immunogenic, consistent with earlier studies. The investigators suggested that the vaccine failed to prevent anal HPV infection because the population had been highly exposed to HPV, and the new infections may have been present at very low levels at baseline. The prevention of oral HPV infections is an important finding, as there are currently no accepted strategies for prevention of HPV-related oropharyngeal cancer.

Borges and colleagues presented data on the occurrence of cancer in the START study (Abstract 160). In the START trial, antiretroviral therapy–naive participants with CD4+ cell counts above 500/µL were randomly assigned to immediate initiation of antiretroviral therapy or delay of therapy until their CD4+ cell count declined to 350/µL or until there was another indication for antiretroviral therapy. Among participants, the overall rate of cancer decreased by 64% (14 vs 39 cancers, respectively; HR, 0.36; 95% CI, 0.19-0.66; P = .001), and risk reduction was greatest for infection-related cancer (6 vs 23, respectively; HR, 0.26; 95% CI, 0.11-0.64; P = .003) (cancers were predominantly Kaposi sarcoma and lymphoma). There was a nonsignificant decrease in cancer unrelated to HIV infection (8 vs 16, respectively; HR, 0.49; 95% CI, 0.21-1.15; P = .1). The effect of early antiretroviral therapy on HIV infection–related cancer did not appear to be mediated by suppression of plasma HIV-1 RNA, whereas viral suppression did mediate the effect of antiretroviral therapy on cancer unrelated to HIV infection. The investigators suggested that other factors such as reduced inflammation or a direct effect of antiretroviral therapy on viral coinfections were responsible for the reduced risk of cancer related to HIV infection.

Recent guidelines for the prevention of cervical cancer have advocated less frequent screening.[10] Silverberg and colleagues conducted a retrospective review of cervical HSILs or cancer in a large health system database (Abstract 162). The investigators conducted a large case-control study to examine the risk factors for these events, including HIV infection. HIV-infected women were 2.3 times more likely to have cervical HSILs or cancer than uninfected women. However, HIV-infected women with CD4+ cell counts above 500/µL had similar risk to that of uninfected women. This suggests that HIV-infected women with high CD4+ cell counts may not require more intensive cervical cancer screening.

Opportunistic Infections

Strategies to Reduce Tuberculosis-Associated Mortality Among Persons With HIV Infection

Tuberculosis (TB) remains the leading killer of persons living with HIV infection worldwide. Delays in TB diagnosis and treatment contribute to this mortality. Two randomized studies sought to show that new TB diagnosis and treatment strategies could reduce this mortality. Novel strategies in both studies failed to reduce mortality. Notably, both studies included timely initiation of antiretroviral therapy. The results were disappointing, somewhat surprising, and highlight the need for better strategies for individuals who present to care with low CD4+ cell counts and the priority of initiating antiretroviral therapy earlier in the course of HIV disease, when TB is much less frequent and less fatal.

The TB Fast Track study randomly assigned 24 clinics in South Africa to 1 of 2 arms: a treatment using a "fast-track" TB clinical algorithm or the standard of care (Abstract 155). Adults with CD4+ cell counts below 150/µL were eligible for participation. In the fast-track arm, participants were categorized as having high, medium, or low risk for TB based on clinical characteristics associated with TB (reduced hemoglobin level and body mass index) and results of a rapid urine lipoarabinomannan (LAM) test. Participants at high risk for TB received immediate TB treatment followed by antiretroviral therapy after 2 weeks. Participants with medium risk had further diagnostic tests, and participants with low risk initiated antiretroviral therapy. Of participants, 45.7% were classified as high risk, 31.5% as medium risk, and 22.8% as low risk. Mortality at 6 months, for the 3030 participants enrolled in the study, did not differ between the 2 arms: 19.0 per 100 person-years (fast track) and 21.5 per 100 person-years (standard of care) (adjusted risk ratio, 0.87; 95% CI, 0.61, 1.24). Antiretroviral therapy was delayed in the intervention arm despite the intent of the study to accelerate the life-saving interventions of TB and HIV treatments.

A "fast-track" TB treatment algorithm for high-risk individuals failed to reduce mortality in a South African study.

The REMEMBER (Reducing Early Mortality & Morbidity by Empiric TB Treatment) study randomly assigned participants screened for TB based on clinical symptoms and rapid sputum tests to either empiric TB treatment or isoniazid preventive therapy (Abstract 745). The rationale for this study was that current clinical and microbiologic rapid diagnostic assessments are too insensitive to identify individuals with low CD4+ cell counts at the highest risk for TB mortality, and that empiric 4-drug TB therapy could reduce this mortality. Eight hundred fifty participants predominantly from Africa were enrolled, with a median CD4+ cell count of 18/µL. At 1 year, mortality was 7.2% with empiric TB treatment and 8.7% with isoniazid preventive therapy, and the probability of AIDS or death was 19.3% and 15.3%, respectively.

Empiric TB treatment in individuals with low CD4+ cell counts in low-income settings failed to reduce rates of mortality.

TB was significantly more frequent among individuals who received 4-drug TB therapy than those who received isoniazid preventive therapy (5.6% vs 2.4%, respectively; $P = .02$). The investigators postulated poorer adherence to 4-drug TB therapy than to isoniazid preventive therapy as one possible explanation for these results.

To address the possibility that screening of participants by urine LAM test could have affected the study results, Bisson evaluated baseline stored urine LAM samples from 67% of participants in the REMEMBER study (Abstract 747). Urine LAM testing would have identified and excluded 28 additional participants in the study. However, after these participants were excluded in a secondary analysis, the same trends reported in the primary analysis were found.

In a related study on TB mortality, Manabe examined mortality outcomes among 804 HIV-infected adults with a presumptive diagnosis of TB enrolled in a TB diagnostic study that used extensive microbiologic tests for final classification (Abstract 743). Of participants, 78% were hospitalized, and 43% were ultimately classified as having TB infection. Six-month mortality rates were high in those with TB and those without TB (26.4% and 26.6%, respectively). Median CD4+ cell count was higher among those without than with TB (114/μL for those without TB vs 59/μL for those with TB). These data show that individuals with TB-like syndromes but without TB require new strategies to reduce mortality rates.

TB among children is even more challenging to diagnose and treat. In an analysis of the International Epidemiologic Databases to Evaluate AIDS (IeDEA) cohort, Carlucci reported that among 295 children (median age, 5.7 years; interquartile range, 2.0-9.6) treated for TB, 22% had unfavorable outcomes (ie, death, treatment failure, or loss to follow up), and outcomes were similarly poor to those of children with and without confirmed microbiologic TB diagnoses (21%).

Extensively Drug-Resistant TB, Multidrug-Resistant TB, and Meropenem for TB Treatment

Understanding of transmission of extensively drug-resistant (XDR) TB is needed to design strategies to halt its spread. Auld described a social network analysis of XDR TB in KwaZulu-Natal, South Africa, from 2010 to 2014 (Abstract 157). The researchers defined a social network connection or an overlapping hospitalization as an epidemiologic link. Among 404 participants with XDR TB, 58% were women, 77% were infected with HIV, and the median age was 34 years. Epidemiologic links were identified in 287 (71%) participants. Of these, 92% were in the same home and 66% had overlapping hospitalizations. These data indicate that the XDR TB epidemic is being driven by transmissions in households and hospitals. Focusing contact tracing at households and limiting exposure of individuals with XDR TB to uninfected persons in hospitals are strategies supported by these findings.

In a South African study, XDR TB was determined to be driven by transmission in households.

Treatment of XDR and multidrug-resistant (MDR) TB often includes injectable aminoglycosides, which are associated with high rates of adverse events, including ototoxic effects. Individuals with HIV infection also require antiretroviral therapy, which may include TDF, a drug that can cause nephrotoxic effects. Brust reported on toxic effect outcomes among 206 individuals with XDR TB (Abstract 756). Among these individuals, 150 had HIV infection and all were receiving antiretroviral therapy. More than half (56%) of individuals experienced hearing loss (≥ grade 1), and 9% experienced severe hearing loss (≥ grade 3). Forty percent developed hypothyroidism requiring replacement therapy. Electrolyte imbalances were common; renal failure was not reported. Persons with HIV infection taking antiretroviral therapy did not experience more toxic effects than persons without HIV infection. These findings highlight that current XDR TB regimens are associated with clinically significant rates of toxic effects but that individuals with HIV infection who are treated with antiretroviral therapy do not experience worse toxic effects than their uninfected counterparts.

Shin and colleagues evaluated rates of treatment success among 403 adults with MDR TB according to HIV infection and antiretroviral therapy status in Botswana from 2006 to 2013 (Abstract 755). Treatment success (microbiologic cure) was similar between HIV-uninfected (82.3%) and HIV-infected (81.0%) individuals taking antiretroviral therapy. Success rates were only 55% among those with HIV infection who were not taking antiretroviral therapy. Mortality rates were lower among HIV-infected individuals treated with antiretroviral therapy than those not treated, but treatment failure was still unacceptably high in all groups (27.5% among HIV-infected persons not taking antiretroviral therapy, 19.2% among HIV-infected persons taking antiretroviral therapy, and 13.2% among uninfected persons).

The high rates of mortality, treatment failure, and toxic effects associated with treatment of XDR and MDR TB in HIV-infected and uninfected persons call attention to the urgent need for new TB treatments. Diacon presented the results of a 2-week intensive microbiologic study examining meropenem and faropenem (Abstract 158LB). The trial tested the hypothesis that meropenem 2 g given intravenously every 8 hours or faropenem 600 mg given orally every 8 hours, each in conjunction with a β-lactamase inhibitor (clavulanic acid), could achieve sufficient plasma levels to inhibit *M. tuberculosis* replication. Because clavulanic acid was not available as a sole agent, it was provided in a coformulated preparation with amoxicillin. In the control (standard TB therapy) and meropenem groups, viable microbiologic load in sputum at 2 weeks was lowered by 0.17 $\log_{10}$

Meropenem plus amoxicillin coformulated with clavulanic acid showed microbiologic activity against TB in an intensive 2-week study.

colony-forming units (CFU)/mL and 0.11 $\log_{10}$ CFU/mL, respectively. There was no reduction observed in the individuals in the faropenem arm, presumably because of the low levels of faropenem achieved with the oral preparation. Most toxic effects observed were gastrointestinal symptoms, attributed by the investigators to amoxicillin and clavulanic acid. These data show proof of principle that meropenem given at this dose with amoxicillin and clavulanic acid can reduce TB levels in 2 weeks, in the range of other potent TB regimens. Identifying carbapenems with activity against TB that can achieve therapeutic levels with oral dosing should be a high priority in the development of TB treatments.

Population-Level Screening for TB

TB symptoms are often present months before an individual presents to a health facility. In the context of the large ongoing PopART (Population Effects of Antiretroviral Therapy to Reduce HIV Transmission) study, an HIV test-and-treat study being conducted in South Africa and Zambia, investigators reported on the uptake and yield of adding TB screening to household HIV testing (Abstract 156). Study staff obtained consent from household members for TB screening at the time of household HIV testing. Sputum was collected for microbiologic diagnosis among persons with TB symptoms. The investigators performed TB screening for 209,429 of 212,819 consenting household members. Of these, 2538 reported TB symptoms, and 1918 (79.3%) had TB test results; 167 of 1918 (8.7%) had a positive TB test result. The investigators concluded that home screening identified TB in nearly 9% of the population and was effective in reaching these individuals before they presented to a health center.

In a household-based TB screening study conducted in 8 communities (103,000 persons) in Haiti, many undiagnosed cases of TB were identified (Abstract 739). Community health workers identified 6926 individuals with suspected TB (7% of the population). Chronic cough was confirmed at a physician visit in 3397 (49%) individuals. Of the 3147 (93%) persons who received HIV and TB microbiologic tests, 302 were HIV infected; 90 (30%) of the 302 HIV-infected persons were diagnosed with TB. Of HIV-uninfected persons, 22% were found to have TB. In this area with high HIV and TB prevalence, household screening and testing were highly effective in identifying undiagnosed cases of these infections.

In Haiti, household screening and testing were highly effective in identifying undiagnosed cases of HIV and TB infections.

In a second analysis from Haiti, Rivera and colleagues evaluated the addition of clinical and microbiologic screening (acid-fast bacilli smear, Xpert MTB/RIF assay, and TB culture) to a routine HIV testing program (Abstract 750). Among 30,316 persons presenting for HIV testing, 3252 (11%) were HIV infected. Among these, 1081 (33%) reported a cough. Among those who reported a cough, 245 (23%) were diagnosed with TB. Sixty-seven percent of the TB diagnoses were confirmed microbiologically. Testing with the Xpert MTB/RIF assay, a rapid combined TB and resistance to rifampicin assay, increased the bacteriologically confirmed diagnoses of TB by 30%. These data illustrate the opportunity for rapid TB diagnosis among persons presenting for HIV testing and the increased yield that may be achieved by incorporating testing with the Xpert MTB/RIF assay.

Cryptococcal Disease

Identifying and treating cryptococcal disease in severely immunosuppressed HIV-infected individuals before they develop clinical meningitis, in populations with a high prevalence of cryptococcal antigenemia, is now recommended as one approach by the World Health Organization.[11] Longly described the outcome of practitioner-initiated screening for cryptococcal antigen (CrAg) among individuals with CD4+ cell counts below 100/µL in Cape Town, South Africa, from 2012 to 2013 (Abstract 759). The protocol called for treatment with oral fluconazole for asymptomatic individuals who tested positive for CrAg. Only 1170 (26.6%) of 4395 eligible individuals were screened for CrAg. The prevalence of cryptococcal antigenemia among screened individuals was 2.1% (24/1170). Information on use of fluconazole was available for 13 of the 24 individuals with cryptococcal antigenemia. Among these, 9 of 13 received fluconazole and none developed cryptococcal meningitis. During the observation period, there were 9 cases of disseminated cryptococcal disease, many of which in individuals who had delays in initiation of antiretroviral therapy. Antiretroviral therapy was initiated for 72% of persons screened and only 48% of persons not screened for CrAg. The investigators appropriately concluded that an implementation approach that relied on practitioner-initiated screening for CrAg without training or feedback was unsuccessful. These results also illustrate that delays in initiation of antiretroviral therapy among persons with low CD4+ cell counts remain a persistent issue.

In a related abstract, Letang reported a more successful approach to CrAg screening (Abstract 760). At the St. Francis referral hospital in Tanzania, "reflex" CrAg screening was performed on all inpatients and outpatients with CD4+ cell counts below 150/µL from 2013 to 2015. Five hundred patients were screened. The prevalence of CrAg was 12% among inpatients and 5.3% among outpatients. Among the cases in which CrAg was identified, a lumbar puncture was performed within 1 day for 31 of 32 individuals; 39% (12/31) had microbiologic evidence of cryptococcal meningitis. Interestingly, 2 of the 12 individuals with evidence of cryptococcal meningitis had no reported neurologic symptoms. The mortality rate of those with cryptococcal meningitis was 86% over the study time period. Mortality among other individuals who tested positive for CrAg (81% were treated with fluconazole) was similar to that among those who tested negative for CrAg (9%).

Predictors of progression from the time of CrAg detection to onset of cryptococcal meningitis are poorly understood.

In the context of an ongoing study on the use of fluconazole for individuals with CrAg, Morawski presented data on outcomes for individuals with detectable CrAg (Abstract 159). Individuals with CrAg and no signs or symptoms of meningitis were treated with fluconazole 800 mg for 2 weeks, followed by fluconazole 400 mg for 8 weeks. In this study population of 151 persons, the median CrAg titer was 1:40. Death occurred in 13.9% of the cohort; 23.2% of participants met the primary end point of death or cryptococcal meningitis. The CrAg titer was highly associated with progression to the primary end point, with a 2.5 greater risk for meningitis when the CrAg titer was greater than 1:160. A CD4+ cell count below 50/μL was the most powerful predictor of outcome. These data advance the understanding of cryptococcal disease progression in the face of a punctuated treatment with fluconazole. These data also raise many additional questions regarding the optimal clinical approach to treatment of individuals with CrAg, including the treatment regimen, treatment duration, and the optimal criteria to tailor treatment for those at the highest risk.

Whether the presence of CrAg without meningitis is associated with neurocognitive changes is unknown. Montgomery and colleagues evaluated neurocognitive function among individuals in African cohorts in 3 groups: 1) those with cryptococcal meningitis (n = 90); 2) those in whom cryptococcal antigenemia was detected (n = 87), with no meningitis; and 3) those with no cryptococcal disease (n = 125) (Abstract 761). The researchers performed a standardized neurocognitive function evaluation at the time of the initiation of antiretroviral therapy and then 4 weeks after the initiation of treatment with fluconazole in group 2. Median CD4+ cell counts and Karnofsky scores among groups 1, 2, and 3, respectively, were 17/μL and 60, 26/μL and 70, and 223/μL and 90. Composite neurocognitive function was lowest among individuals with cryptococcal meningitis, and it was lower among individuals with CrAg only than among HIV-infected individuals with no cryptococcal disease; however, CD4+ cell counts were different among these groups. Neurocognitive function in the cohort of 87 persons with CrAg but without meningitis improved to within 1 standard deviation of persons without cryptococcal disease, 4 weeks after treatment with fluconazole and antiretroviral therapy. It is difficult to assess whether this improvement was attributable to fluconazole, antiretroviral therapy, or both.

Boulware and colleagues conducted an analysis of individuals with cryptococcal disease to determine if clinical and immune parameters could help distinguish immune reconstitution inflammatory syndrome (IRIS) from disease relapse (Abstract 762). The investigators applied a standard definition of IRIS and used cultures from cerebrospinal fluid to define relapse. Among the cohort analyzed, 70 persons had 75 episodes of recurrent meningitis, 62 of which were classified as IRIS and 13 of which were classified as disease relapse. None of clinical lab parameters (CD4+ cell count, HIV RNA level) examined distinguished between these 2 clinical scenarios, which call for different treatment approaches. Interleukin (IL)-13 (a cytokine associated with uncontrolled cryptococcal disease in murine models) in cerebrospinal fluid was 35-fold higher in those with disease relapse than in those with IRIS. Other inflammatory markers (interferon-γ, IL-4, and IL-17) were higher in those with IRIS than those with disease relapse. Without fungal cultures, it remains difficult to distinguish between IRIS and relapse of cryptococcal disease. The investigators noted that the best way to reduce disease relapse is optimal treatment of cryptococcal meningitis. Many places in Africa still treat cryptococcal meningitis with fluconazole alone because of a lack of access to amphotericin B.

All cited abstracts appear in the CROI 2016 Abstracts eBook, available online at www.CROIconference.org.

Acknowledgement: The authors would like to thank Timothy J. Wilkin, MD, MPH, for contributing the section on cancer.

Financial affiliations in the past 12 months: Drs Havlir and Currier have no relevant financial affiliations to disclose.

Additional References Cited in Text

1. Serrano-Villar S, Sainz T, Lee SA, et al. HIV-infected individuals with low CD4/CD8 ratio despite effective antiretroviral therapy exhibit altered T cell subsets, heightened CD8+ T cell activation, and increased risk of non-AIDS morbidity and mortality. *PLoS Pathog*. 2014;10(5):e1004078.
2. Goff DC, Jr., Lloyd-Jones DM, Bennett G, et al. 2013 ACC/AHA guideline on the assessment of cardiovascular risk: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. *J Am Coll Cardiol*. 2014;63(25 Pt B):2935-2959.
3. Grundy SM, Cleeman JI, Merz CN, et al. Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel III guidelines. *Circulation*. 2004;110(2):227-239.
4. Wilson EM, Singh A, Hullsiek KH, et al. Monocyte-activation phenotypes are associated with biomarkers of inflammation and coagulation in chronic HIV infection. *J Infect Dis*. 2014;210(9):1396-1406.
5. Barbour JD, Jalbert EC, Chow DC, et al. Reduced CD14 expression on classical monocytes and vascular endothelial adhesion markers independently associate with carotid artery intima media thickness in chronically HIV-1 infected adults on virologically suppressive anti-retroviral therapy. *Atherosclerosis*. 2014;232(1):52-58.
6. Westhorpe CL, Maisa A, Spelman T, et al. Associations between surface markers on blood monocytes and carotid atherosclerosis in HIV-positive individuals. *Immunol Cell Biol*. 2014;92(2):133-138.
7. Ogilvie RP, Everson-Rose SA, Longstreth WT, Jr., Rodriguez CJ, Diez-Roux AV, Lutsey PL. Psychosocial factors and risk of incident heart failure: the multi-ethnic study of atherosclerosis. *Circ Heart Fail*. 2016;9(1):e002243.
8. Mulligan K, Glidden DV, Anderson PL, et al. Effects of emtricitabine/tenofovir on bone mineral density in HIV-negative persons in a randomized, double-blind, placebo-controlled trial. *Clin Infect Dis*. 2015;61(4):572-580.
9. Damouche A, Lazure T, Avettand-Fenoel V, et al. Adipose tissue is a neglected viral reservoir and an inflammatory site during chronic HIV and SIV infection. *PLoS Pathog*. 2015;11(9):e1005153.
10. Panel on Opportunistic Infections in HIV-Infected Adults and Adolescents. Guidelines for the prevention and treatment of opportunistic infections in HIV-infected adults and adolescents: recommendations from the Centers for Disease Control and Prevention, the National Institutes of Health, and the HIV Medicine Association of the Infectious Diseases Society of America. http://aidsinfo.nih.gov/contentfiles/Adult_OI.pdf. Accessed on May 31, 2016.
11. World Health Organization. Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection. Recommendations for a public health approach. http://apps.who.int/iris/bitstream/10665/85321/1/9789241505727_eng.pdf. Accessed on May 31, 2016.

